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Keywords

Ostrinia nubilalis, monoterpenoids, natural products

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Influence of Dietary Applied Monoterpenoids and Derivatives on Survival and Growth of the European Corn Borer (Lepidoptera: Pyralidae)

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ABSTRACT Sixteen natural monoterpenoids and 6 synthetic derivatives were selected for study of larvicidal activity and growth inhibitory effect against the European corn borer, *Ostrinia nubilalis* (Hübner). For this study, 2 different dietary exposure bioassays were used: compounds applied on the diet surface (on-diet), and compounds incorporated into the diet (in-diet). Most of the monoterpenoid compounds showed some degree of larvicidal activity in both bioassay procedures after a 6-d exposure period. Among the monoterpenoids, pulegone was the most active. Larvicidal toxicities were significantly enhanced for the structurally modified compounds; monoterpenoid derivatives MTEE-25 (2-fluoroethyl thymyl ether) and MTEE-P (propargyl citronellate) were most toxic to borer larvae. When reared on diet containing the monoterpenoids or their derivatives, changes in developmental parameters and pupal weights of the European corn borer also were noted when they were fed several of the compounds. Some larvae reared on treated diet with higher concentrations of the test compounds died before pupating. In general, growth and development of the European corn borer were affected by monoterpenoid compounds, and some compounds such as *l*-menthol, pulegone, MTEE-25, and MTEE-P acted as insect growth inhibitors.

KEY WORDS *Ostrinia nubilalis*, monoterpenoids, natural products

THE EUROPEAN CORN borer, *Ostrinia nubilalis* (Hübner), is a serious pest of corn and other cultivated plants in the northern hemisphere, including Europe, Asia, and North America. Since this insect was introduced into the United States >70 yr ago, it has rapidly become one of the most important insect pests of corn because of its ability to adapt to the new environment and to attack to a large number of host plants (Beck 1989, Hudon et al. 1989, Ellsworth and Bradley 1992). From the early years of European corn borer research, chemical insecticides have played a major role in crop protection because of the high economic value of field corn, popcorn, and sweet corn for the fresh market (Thompson and White 1977, Grafius et al. 1990, Weissling et al. 1992, Hutchison 1993, Rinkleff 1995). However, the use of synthetic insecticides on corn fields can lead to several potentially adverse effects, including water and soil contamination, insect resistance, and toxicity to nontarget species. Currently, integrated pest management (IPM) is receiving increased attention for European corn borer control (Whitford et al. 1987, Ding et al. 1989). Research on insect growth regulators is a potential pest management tool, and some insect growth regulators have been proved toxic against several lepidopteran larvae

(Chandler et al. 1992, Haynes and Smith 1993, Chandler 1994).

Plant-derived compounds have various useful biological properties (Pillmoor et al. 1993) and more diversified modes of action on various target systems (Mikolajczak et al. 1984, Harborne 1989, Houseman et al. 1992). Monoterpenoids, plant secondary metabolites found in the essential oils of higher plants, are successful examples, and they are believed to aid plants as chemical defenses against phytophagous organisms in nature. Many monoterpenoids can act as insect repellents, attractants, oviposition cues, and antifeedants as well as killing agents in numerous insects (Brattsten 1983, Duke 1991). Karr and Coats (1992) reported the influence of some monoterpenoids on the growth and development of nymphs of German cockroach, *Blattella germanica* (L.). Beninger et al. (1993) showed that the diterpene 3-epicaryoptin reduced growth and increased mortality of the European corn borer larvae when incorporated into artificial diet, and that pupal deformities and time to pupation increased. Pulegone-containing diet (either 0.01 or 0.1%) retarded development and inhibited reproduction of last-instar southern armyworms, *Spodoptera eridania* (Cramer) (Gunderson et al. 1985), and menthol reduced growth and inhibited pupation of the variegated cutworm, *Peridroma sausta* (Hübner) (Harwood et al. 1990).

Many researchers studied insect growth regulators, monoterpenoids, and the European corn borer sepa-

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rately; however, little information is available on the effects of monoterpenoids as insect growth regulators on the European corn borer. Our objective was to determine if several monoterpenoids and their derivatives influence the survival, growth and development, pupation rate, adult emergence rate, pupal weight, and duration of larval and pupal growth periods of European corn borer larvae. We also observed the differences in results from 2 different application methods—the test compounds applied either onto the surface of the diet or incorporated into the diet.

Materials and Methods

Chemicals and Insects. Sixteen natural monoterpenoids showing insecticidal properties (unpublished data) and other chemical reagents used in this study were obtained from Aldrich (Milwaukee, WI), Pfaltz and Bauer (Waterbury, CT), and Sigma (St. Louis, MO). Also, 20% (AI) pyrethrins (Pet Chemicals, Miami Springs, FL) and rotenone (Aldrich) were obtained and used as standards. All solvents and chemicals were of certified grade or >95% purity. They were used directly for bioassays to determine larvicidal toxicity and insect growth inhibitory effects, and some were used as starting materials for the derivatization of monoterpenoids with chemical reagents in this laboratory. Details on the derivatization and identification have been reported previously (Tsao et al. 1995). All monoterpenoids and their derivatives used in this study are listed in Figs. 1 and 2, respectively. For this bioassay, the European corn borer was chosen because of its economic importance and convenience of rearing and supply. European corn borer egg masses provided newly hatched larvae for this study. The egg masses were obtained over a 24-h period from laboratory colonies reared on an artificial diet (Reed et al. 1972, Freedman 1979) from the Corn Insects Research Unit, USDA-ARS, Iowa State University, Ames, and placed in an open transparent plastic bag. Egg masses were maintained under incubation conditions of $25 \pm 1^\circ\text{C}$, 40–60% RH, and a photoperiod of 14:10 (L:D) h, and were observed daily until the proper hatching time when the black larval head capsule became clearly visible. The plastic bag was then closed to prevent larvae from escaping and newly hatched larvae were ready for testing. Late 2nd instars also were obtained from the same laboratory for comparison with the results from the experiment with newly hatched larvae.

Bioassays and Statistical Analysis. Two different chemical application methods were conducted. Dripping the solution with a pasteur pipette onto the solidified artificial diet (on-diet test) or mixing the solution in the diet before it solidified (in-diet test). All ingredients of artificial diet for European corn borer larvae were supplied by the USDA-ARS Corn Insects Research Unit, Ames, IA.

On-Diet Test. After dispensing ≈ 8 ml of prepared diet ($\approx 58^\circ\text{C}$) with a plastic squeeze bottle (200 ml) into clear plastic cups (29 ml, Creamer, Prod. No. 9051, Holton Industries, Frenchtown, NJ) held on a cup tray

(30 cups in a tray, Prod. No. 9040, Holton), the dietary preparations were allowed to solidify at room temperature ($25 \pm 2^\circ\text{C}$). The trays with cups were stacked and packaged in heavy plastic bags tagged with names and dates and stored at 4°C until they were used. Diets from the cold storage room were warmed to room temperature before use. Serial dilutions of each monoterpenoid and derivative were prepared by using certified acetone before treatment. The appropriate amounts from $0.2 \mu\text{g}$ to 20 mg of compound in $200 \mu\text{l}$ of acetone solution were applied with an Eppendorf pipette to the surface of the diet (≈ 3 cm diameter), and the cups were open for 1 h to allow the solvent to evaporate under ambient conditions. One neonate European corn borer larva was transferred onto the treated surface of each diet with a fine paint brush, and the cups were covered with lids lined with Saran. One treatment group consisted of 5 cups, and each treatment group was replicated 4 or 5 times. Untreated diet and the same aliquot of acetone-treated diet also were used for controls in this test. All prepared materials were kept at room temperature ($25 \pm 2^\circ\text{C}$), and larvae were observed daily. Larval mortalities were assessed at 6 d after treatment by counting dead European corn borer, and mortality was calibrated using the Abbott formula (Abbott 1925) with control data. Larvicidal activities of monoterpenoids and derivatives against the 1st-instar European corn borers were expressed as LC_{50} values (milligrams per cup) by using probit analysis (SAS Institute 1991). In addition, growth of surviving larvae was measured and recorded daily up to adulthood for the following growth parameters: pupal weights, duration of the larval period until pupation, and duration of the pupal period until emergence. Numbers of pupae and adults, weight (grams), and duration periods (days) were expressed as means \pm SE and compared with controls. Least significant differences at $P = 0.05$ (SAS Institute 1991) were used to separate the means.

In-Diet Test. To determine any differences between the 2 chemical application methods, the following test was designed. Monoterpenoids were mixed with the artificial diet before the diet solidified. Serial dilutions of chemical compounds were prepared as previously described with certified acetone before the test. The temperature of the artificial diet carried in a warm jar was maintained at $\approx 58^\circ\text{C}$ with running hot water to prevent hardening. Keeping the optimum temperature is very important because higher temperatures can degrade some ingredients such as vitamins, and low temperatures can allow the medium to solidify too quickly. Prepared chemical solutions were incorporated when the dietary preparations, in squeeze bottles, cooled down to $\approx 40^\circ\text{C}$; the mixtures were then thoroughly blended. Maximum acetone volume allowed was up to 2.5 ml in 150 ml of diet. Treatment concentrations ranged from 0.001 to 10,000 ppm (by weight) in the diet mixtures. The treated diet mixtures were poured into plastic cups in a holder tray described earlier and were allowed to harden at room temperature. The cups were kept open for 6 h to allow the organic solvent to evaporate under ambient con-

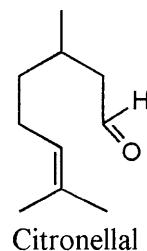
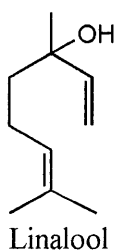
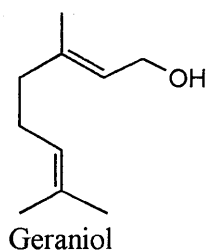
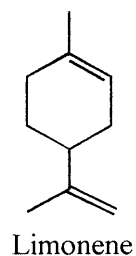
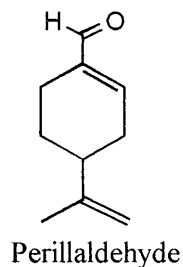
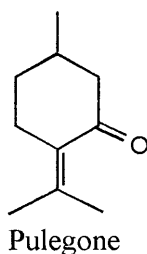
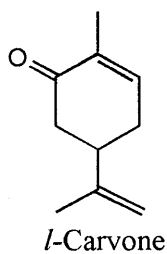
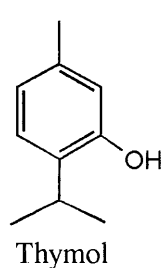
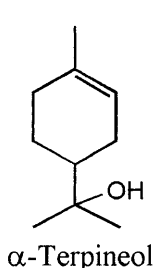
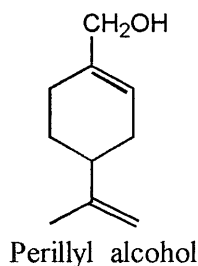
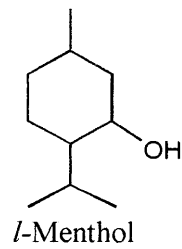
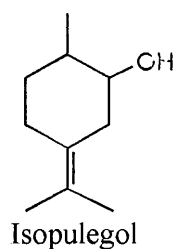
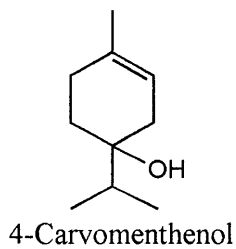
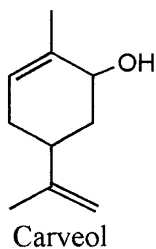
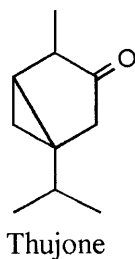
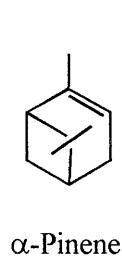
Acyclic**Monocyclic****Bicyclic**

Fig. 1. Monoterpenoid structures.

ditions, packed in a heavy plastic bag, and stored in a cold room at 4°C until used (the next day). One neonate larva was placed individually on the center of each diet treated with a monoterpene at various

concentrations; each cup was then covered with a lid lined with Saran. Five cups made 1 experimental unit, and each unit was replicated 6 times. Untreated and acetone-only treated diet also were used as controls,

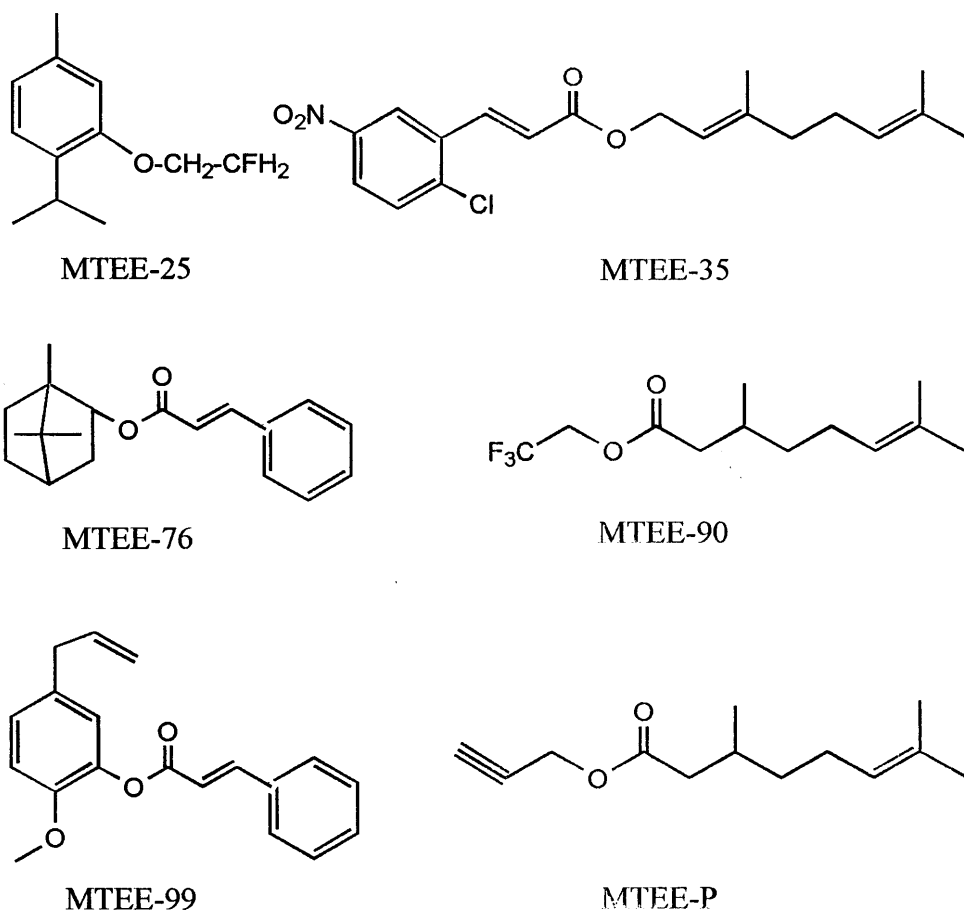


Fig. 2. Monoterpenoid derivative structures: MTEE-25, 2-fluoroethyl thymyl ether; MTEE-35, geranyl 2-chloro-5-nitrocinnamic acid ester; MTEE-76, bornyl cinnamic acid ester; MTEE-90, 2,2,2-trifluoroethyl citronellal acid ester; MTEE-99, eugenyl cinnamic acid ester; MTEE-P, propargyl citronellate.

and a different set of solvent tests was run concurrently for determining the safety level of 4 solvents (acetone, methyl alcohol, ethyl alcohol, and ethyl acetate) on the European corn borer larvae at different concentrations (0.05, 0.5, 2.5, and 5% in each medium). The European corn borer larvae were kept at $25 \pm 2^\circ\text{C}$ and were observed daily for the determination of larval growth, mortality, and the appearance of pupae and adults. Each pupa was weighed, and the duration of larval period and pupal period also were recorded. Larval mortalities were assessed at 6 d after treatment, and data were adjusted using the Abbott formula (Abbott 1925) with control data. Larvicidal toxicities of monoterpenoids against 1st-instar European corn borers reared on the artificial diet incorporated with these compounds were expressed by estimated LC_{50} values as described previously. In addition, an experiment with late 2nd instars was conducted with the prepared diet mixed with compounds in comparison with the result from the test conducted with the neonate European corn borer larvae. Data were collected and

analyzed in the same way, using LSDs and probit analysis.

Results

Larvicidal Toxicity. Most monoterpenoids and their derivatives selected for this study showed some degree of larvicidal activity in both administrative routes, when they were applied on the solidified diet surface (on-diet test) or when they were incorporated into the diet (in-diet test). Generally, the larvicidal effects occurred mostly between 3 and 6 d after treatments, and the mortality data from daily observations were accumulated until the 6th d. The dead insect bodies appeared dark and liquidified. Significant differences occurred among monoterpenoids and derivatives according to LC_{50} values and 95% CL.

On-Diet Test. Larval mortality of 1st instars was moderately high at 6 d when the selected monoterpenoids and derivatives were applied on the surface of the solidified diet (Table 1). Within the group of 16

Table 1. Larvicidal toxicity (6-d mortality) against 1st-instar *O. nubilalis* of some selected monoterpenoids and derivatives applied on the diet

Compound	Doses	<i>n</i> ^a	Slope ± SE	LC ₅₀ 95% CL ^b (mg per cup)	
Monoterpenoids					
Carveol	6	199	—	>20.0	—*
4-Carvomenthenol	6	199	—	>20.0	—
<i>l</i> -Carvone	6	199	−0.94 ± 0.15	3.69	2.55–5.56
Citronellal	6	199	−0.83 ± 0.11	17.6	6.96–63.7
Geraniol	6	199	−0.54 ± 0.13	2.43	1.62–3.80
Isopulegol	6	199	−1.78 ± 0.19	10.5	7.08–16.8
Limonene	6	199	—	>20	—
Linalool	6	199	—	>20.0	—
<i>l</i> -Menthol	6	199	−0.55 ± 0.13	2.35	1.59–3.60
Perillaldehyde	6	199	−0.10 ± 0.11	18.5	8.20–56.6
Perillyl alcohol	5	179	—	>2.00	—
α-Pinene	6	199	—	>20.0	—
Pulegone	6	199	0.99 ± 0.16	0.29	0.20–0.40
α-Terpineol	6	199	—	>20.0	—
Thujone	6	199	−2.44 ± 0.30	5.40	4.17–7.07
Thymol	6	199	−0.31 ± 0.13	1.62	1.10–2.46
Derivatives					
MTEE-25 ^c	5	179	0.57 ± 0.16	0.27	0.16–0.50
MTEE-35	5	179	—	>2.00	—
MTEE-76	5	179	0.72 ± 0.16	0.18	0.11–0.33
MTEE-90	6	199	−0.29 ± 0.11	2.01	1.18–3.69
MTEE-99	5	179	—	>2.00	—
MTEE-P	6	199	3.01 ± 0.30	0.05	0.04–0.07
Standards					
Pyrethrins ^d	6	199	104 ± NA	0.0002	NA
Rotenone	6	199	4.24 ± 0.43	0.03	0.02–0.04

^a Number of *O. nubilalis* larvae tested.

^b LC₅₀s are means of 4 replicates; LC₅₀ values were determined by probit analysis (SAS Institute 1991); concentrations are expressed in mg of compounds on the diet in cups; larvicidal activity is considered significantly different when the 95% CLs fail to overlap. *, Unable to achieve 50% in laboratory tests.

^c MTEE, monoterpenoid derivatives (see Fig. 2).

^d Concentration adjusted for 20% (AI); NA, data not available.

monoterpenoids tested, pulegone, a ketone type monocyclic monoterpenoid, was the most active, followed by thymol, a phenolic monocyclic monoterpenoid; *l*-menthol, an alcoholic monocyclic monoterpenoid; and geraniol, an alcoholic acyclic monoterpenoid. Carveol, 4-carvomenthenol, limonene, linalool, α-pinene, and α-terpineol were the least toxic. In general, monocyclic monoterpenoids were more toxic to the European corn borer larvae than acyclic and bicyclic monoterpenoids. The larvicidal toxicities of the monoterpenoid ether and ester derivatives (MTEEs) were enhanced significantly compared with the other derivatives. Among the derivatives, MTEE-P (propargyl citronellate) was the most enhanced. MTEE-P also showed an activity that was greater than that of MTEE-90 (2,2,2-trifluoroethyl

citronellate), which has the same parent monoterpenoid. The LC₅₀ value of MTEE-25 (2-fluoroethyl thymyl ether, a fluorinated thymol derivative) was 6 times more active than its parent compound, thymol.

In-Diet Test. Table 2 shows larval mortality produced by 4 organic solvents, commonly used in bioassays, after being added to the diet. Methyl alcohol was the safest solvent to the 1st-instar European corn borers for dietary bioassays, followed by acetone and ethyl alcohol, whereas ethyl acetate was the most toxic to larvae. Larval mortality, when it occurred, was usually within the 1st 4 d.

Table 3 shows the larvicidal toxicity of 4 selected monoterpenoids incorporated in the diet against late 2nd instars. The most toxic monoterpenoids were *l*-menthol (LC₅₀, 17.4 ppm), and pulegone (LC₅₀, 26.3

Table 2. Larval mortality of 1st-instar *O. nubilalis* when 4 organic solvents were incorporated into diet

Solvent	d after treatment	% mortality ± SEM ^a				
		5	2.5	0.5	0.05	Control
Acetone	4 d	100 ± 0a	80.0 ± 0b	20.0 ± 11.6c	6.70 ± 6.67c	10.0 ± 6.32c
Ethyl alcohol	4 d	100 ± 0a	93.3 ± 6.67a	26.7 ± 6.67b	13.3 ± 6.67bc	10.0 ± 6.32c
Methyl alcohol	4 d	73.3 ± 13.3a	33.3 ± 6.67b	20.0 ± 11.6b	6.70 ± 6.67b	10.0 ± 6.32b
Ethyl acetate	4 d	100 ± 0a	100 ± 0a	20.0 ± 11.6b	13.3 ± 6.67b	10.0 ± 6.32b

All values are means ± standard error of mean (SEM) of 4 replicates. Means within a row followed by the same letter are not significantly different (LSD, P = 0.05, SAS Institute 1991).

^a Concentrations are expressed as grams of compounds in 100 g of the diet in cups.

Table 3. Larvicidal toxicity of some selected monoterpenoids incorporated into the diet against 2nd-instar *O. nubilalis* after 6 d

Compound	No. doses	n ^a	Slope \pm SE	LC ₅₀ ^b , ppm	95% CL ^b
Geraniol	5	160	—	>10,000	—*
<i>l</i> -Menthhol	5	160	-1.08 \pm 0.06	17.4	14.4–20.8
Pulegone	5	160	-1.48 \pm 0.06	26.3	22.5–30.6
Rotenone	5	160	-0.93 \pm 0.07	52.3	45.3–60.4

^a Number of *O. nubilalis* larvae tested.

^b LC₅₀s are means of 4 replicates; LC₅₀ values were determined by probit analysis (SAS Institute 1991); concentrations are expressed in parts per million (μ g of compounds per g of diet); 95% CL: larvicidal activity is considered significantly different when the 95% CLs fail to overlap; *, unable to achieve 50% in laboratory tests.

ppm), which was slightly less toxic. The toxicities of both pulegone and *l*-menthol were greater than the standard compound, rotenone (LC₅₀, 52.3 ppm).

Chronic Evaluation Tests. Pupation Rates. Some larvae reared on the monoterpenoid-treated diet at higher concentrations were dead before undergoing pupation (larvicidal toxicity). All larvae died before pupation with the 20-mg treatment of *l*-carvone, *l*-menthol, pulegone, thujone, thymol, MTEE-25, MTEE-90, and MTEE-P. Pulegone, MTEE-25, and MTEE-P killed all larvae even at the 2-mg treatment. Some monoterpenoids reduced percentage pupation rates in a broad range of concentrations (5–85% compared with the control) (Table 4). *l*-Menthhol showed 20–30% reduced pupation at all concentrations from 0.2 μ g to 2 mg), and monoterpenoid derivatives MTEE-25, MTEE-99, and MTEE-P showed a greatly reduced pupation rate (20–70%) at most concentrations.

Similar trends were obtained from the in-diet tests (Table 5 and 6). Pulegone killed all larvae at the higher concentrations (10,000 or 1,000 ppm) and reduced the pupation rate slightly in the in-diet test with 1st instars (Table 5). Pulegone reduced pupation to an even greater degree (38–48%) with 2nd instars (Table 6). *l*-Menthhol also reduced the pupation rates (23–63%).

Emergence Rate. In general, emergence rates were lower than pupation rates, even in controls (Tables 4–6). The number of adults emerging from the pupae was affected slightly by any treatment and the reductions varied from 10 to 45% relative to the number of pupae for treated European corn borer larvae. The differences between pupation rates and emergence rates in the controls were 5.00, 43.7, and 40.0% observed in the on-diet test (reared from 1st instars), the in-diet test (reared from 1st instars), and the in-diet test (reared from 2nd instars), respectively. The compounds most active in Table 4 were *l*-menthol, perillaldehyde, α -pinene, MTEE-25, MTEE-90, and MTEE-99. These compounds were more active than rotenone but not as active as pyrethrins. Among them, MTEE-25 showed a significant reduction (30–35%) relative to the pupation rates in the on-diet test.

Larval and Pupal Period. Durations of larval period were not influenced significantly by monoterpenoid compounds studied in the on-diet and the in-diet (1st instar) assays (Tables 4 and 5); however, the durations

of the larval period of European corn borer reared from 2nd instars in the in-diet test were affected by treatments. Geraniol, *l*-menthol, and pulegone showed significantly longer larval periods in comparison with the duration of the control larval period (Table 6). Durations of the pupal period were not influenced significantly by any treatment.

Pupal weights were influenced by monoterpenoid compound treatments, and most pupal weights were reduced, with some exceptions. In the on-diet test, citronellal, geraniol, isopulegol, linalool, thujone, and MTEE-25 were the monoterpenoid compounds that reduced pupal weight, whereas 4-carvomenthenol increased pupal weights slightly (Table 4).

Discussion

Although many natural monoterpenoids have been studied by researchers and cause a variety of effects in insects (Karr and Coats 1988; Bauske et al. 1994; Rice and Coats 1994 a, b), their spectra of activity and modes of action as regulators of insect growth and development are poorly understood. The current study broadens our understanding of the potential for monoterpenoids as insecticides acting via ingestion. Laboratory dietary bioassays conducted showed that 16 monoterpenoids and 6 derivatives had various biological effects against the European corn borer. Natural monoterpenoids showed larvicidal activities against the European corn borer from moderate to high degrees, and some derivatives demonstrated enhanced larvicidal activity compared with the parent monoterpenoid. Among the monoterpenoids and derivatives tested, pulegone and MTEE-P (propargyl citronellate) were the most active compounds. Monocyclic monoterpenoids were generally more effective than acyclic and bicyclic monoterpenoids in the dietary bioassays. Rice and Coats (1994a) observed a similar trend with regard to larvicidal activity of monoterpenoids against the southern corn rootworm, *Diabrotica undecimpunctata howardi* (Herbst) in a soil bioassay. Several derivatives, including trifluoroacetates of cyclic monoterpenoids, similarly enhanced activity relative to their parent compound (Rice and Coats 1994b). This illustrated that structural modification, especially functional groups, can influence insecticidal effectiveness. They suggested, moreover, that the structural characteristics of monoterpenoids such as shape, saturation, and functional group could influence their insecticidal properties. The on-diet and in-diet tests both showed similar tendencies, although the compounds were administered in 2 different ways. The comparison of 1st and 2nd instars for the in-diet experiments revealed the 1st instars to be more susceptible to rotenone than the 2nd instars, whereas an opposite trend was evident for *l*-menthol (i.e., older larvae [2nd instars] were more susceptible than the neonates). Pulegone toxicity was similar in both experiments (i.e., the 2 stages of European corn borer showed approximately equal susceptibility) (Tables 5 and 6). The example of *l*-menthol was particularly striking in that the late 2nd instars were affected det-

Table 4. Effects of selected monoterpenoids and derivatives applied on the diet on development of *O. nubilalis*, beginning with neonate 1st instars

Compound	<i>n</i> ^a	Development	Concentrations ^b						Control (blank)
			20 mg	2 mg	0.2 mg	0.02 mg	2 μg	0.2 μg	
Carveol	199	% pupation	35.0 ± 9.57c	75.0 ± 5.00b	95.0 ± 5.00a	85.0 ± 9.57a	85.0 ± 9.57a	100 ± 9.57a	90.0 ± 10.0a
		% emergence	10.0 ± 5.77c	50.0 ± 10.00b	75.0 ± 5.00ab	70.0 ± 19.1ab	80.0 ± 14.1a	90.0 ± 5.77a	85.0 ± 5.00a
		Larval period, d	30.3 ± 2.62a	25.7 ± 1.45a	25.8 ± 1.28a	25.6 ± 1.49a	24.3 ± 1.00a	24.5 ± 0.77a	26.3 ± 0.75a
		Pupal wt, mg	73.6 ± 3.63a	82.9 ± 0.50a	80.2 ± 3.26a	76.1 ± 3.49a	85.2 ± 4.83a	82.1 ± 4.20a	82.1 ± 2.37a
4-Carvomenthenol	199	% pupation	60.0 ± 11.5b	75.0 ± 9.57ab	80.0 ± 8.16a	95.0 ± 5.00a	85.0 ± 9.57a	95.0 ± 5.00a	90.0 ± 10.0a
		% emergence	40.0 ± 8.17b	60.0 ± 8.17ab	70.0 ± 12.9a	70.0 ± 12.9a	65.0 ± 15.0ab	80.0 ± 8.17a	85.0 ± 5.00a
		Larval period, d	26.4 ± 1.22a	28.4 ± 1.30a	25.8 ± 1.03a	27.6 ± 1.21a	24.7 ± 0.89a	24.5 ± 0.95a	26.3 ± 0.75a
		Pupal wt, mg	70.7 ± 4.04b	95.8 ± 5.09a	83.3 ± 4.50b	78.5 ± 4.08ab	87.4 ± 3.14a	92.9 ± 3.56a	82.1 ± 2.37ab
<i>l</i> -Carvone	199	% pupation	0 ± 0c	75.0 ± 5.00b	75.0 ± 9.57b	90.0 ± 10.0a	90.0 ± 5.77a	95.0 ± 5.00a	90.0 ± 10.0a
		% emergence	0 ± 0d	55.0 ± 5.00c	70.0 ± 12.9b	75.0 ± 12.6b	90.0 ± 5.77a	85.0 ± 5.00a	85.0 ± 5.00a
		Larval period, d	—	27.5 ± 1.40a	24.7 ± 0.67a	27.1 ± 1.02a	24.3 ± 0.64a	23.3 ± 0.53a	26.3 ± 0.75a
		Pupal wt, mg	—	83.3 ± 6.56a	81.8 ± 3.51a	81.2 ± 4.61a	76.8 ± 5.19ab	65.5 ± 5.35b	82.1 ± 2.37a
Citronellal	199	% pupation	15.0 ± 5.00c	80.0 ± 8.16a	90.0 ± 5.77a	80.0 ± 8.16a	85.0 ± 15.0a	70.0 ± 12.9b	90.0 ± 10.0a
		% emergence	15.0 ± 5.00c	70.0 ± 5.77ab	65.0 ± 12.6ab	65.0 ± 15.0ab	75.0 ± 12.6a	55.0 ± 17.1b	85.0 ± 5.00a
		Larval period, d	26.0 ± 1.17a	26.4 ± 0.85a	24.4 ± 0.70a	25.0 ± 0.91a	24.9 ± 0.77a	23.1 ± 0.91a	26.3 ± 0.75a
		Pupal wt, mg	77.0 ± 7.85a	68.5 ± 3.58ab	64.0 ± 5.54b	59.9 ± 4.30b	63.3 ± 5.43b	56.6 ± 6.66b	82.1 ± 2.37a
Geraniol	199	% pupation	5.00 ± 5.00c	60.0 ± 11.5b	85.0 ± 5.00a	85.0 ± 9.57a	90.0 ± 10.0a	90.0 ± 5.77a	90.0 ± 10.0a
		% emergence	0 ± 0c	50.0 ± 12.9b	65.0 ± 5.00b	60.0 ± 11.5b	80.0 ± 8.16a	45.0 ± 15.0b	85.0 ± 5.00a
		Larval period, d	29.0 ± 0a	27.9 ± 1.35a	23.7 ± 0.76a	25.8 ± 1.09a	24.4 ± 0.80a	25.8 ± 1.17a	26.3 ± 0.75a
		Pupal wt, mg	37.0 ± 0c	73.5 ± 6.87ab	61.0 ± 4.75b	70.5 ± 6.38ab	67.4 ± 4.64ab	58.4 ± 5.04b	82.1 ± 2.37a
Isopulegol	199	% pupation	15.0 ± 5.00b	80.0 ± 8.16a	85.0 ± 9.57a	95.0 ± 5.00a	85.0 ± 5.00a	80.0 ± 8.16a	90.0 ± 10.0a
		% emergence	0 ± 0b	75.0 ± 5.00a	70.0 ± 5.77a	80.0 ± 8.16a	70.0 ± 5.77a	70.0 ± 17.3a	85.0 ± 5.00a
		Larval period, d	30.3 ± 1.36a	24.7 ± 0.57b	24.7 ± 0.99b	24.5 ± 0.86b	23.2 ± 0.54b	23.9 ± 0.83b	26.3 ± 0.75b
		Pupal wt, mg	95.3 ± 1.39a	69.6 ± 4.7b	57.2 ± 7.18b	64.1 ± 5.71b	57.1 ± 5.34b	62.9 ± 5.02b	82.1 ± 2.37a
Limonene	199	% pupation	75.0 ± 9.57b	90.0 ± 10.0a	85.0 ± 15.0ab	90.0 ± 5.77a	95.0 ± 5.00a	90.0 ± 10.0a	90.0 ± 10.0a
		% emergence	55.0 ± 5.00b	65.0 ± 12.6b	85.0 ± 15.0a	70.0 ± 12.9b	90.0 ± 5.77a	80.0 ± 8.16a	85.0 ± 5.00a
		Larval period, d	26.1 ± 1.28a	27.5 ± 1.03a	23.2 ± 0.63a	27.5 ± 1.49a	25.0 ± 0.74a	25.1 ± 1.09a	26.3 ± 0.75a
		Pupal wt, mg	76.5 ± 6.66a	87.4 ± 4.24a	84.3 ± 4.29a	84.6 ± 6.68a	79.2 ± 3.79a	69.3 ± 4.18a	82.1 ± 2.37a
Linalool	199	% pupation	55.0 ± 15.0c	85.0 ± 5.00b	85.0 ± 9.57b	95.0 ± 5.00a	100 ± 0a	95.0 ± 5.00a	90.0 ± 10.0a
		% emergence	45.0 ± 18.9c	60.0 ± 8.16bc	75.0 ± 9.51a	85.0 ± 9.51a	75.0 ± 9.51a	80.0 ± 11.5a	85.0 ± 5.00a
		Larval period, d	27.9 ± 1.13a	24.2 ± 0.69a	26.0 ± 0.93a	24.8 ± 0.87a	23.6 ± 0.57a	23.9 ± 0.78a	26.3 ± 0.75a
		Pupal wt, mg	78.1 ± 6.00a	67.9 ± 5.00ab	68.6 ± 4.59ab	68.9 ± 3.88ab	60.2 ± 4.70b	57.9 ± 5.46b	82.1 ± 2.37a
<i>l</i> -Menthol	199	% pupation	0 ± 0c	70.0 ± 5.77b	65.0 ± 17.1b	60.0 ± 8.16b	65.0 ± 15.0b	65.0 ± 15.0b	90.0 ± 10.0a
		% emergence	0 ± 0c	40.0 ± 14.1b	45.0 ± 15.0b	55.0 ± 9.51b	55.0 ± 15.0b	50.0 ± 12.9b	85.0 ± 5.00a
		Larval period, d	0 ± 0b	31.5 ± 1.26a	28.9 ± 1.70a	26.2 ± 1.15a	27.3 ± 1.78a	26.5 ± 1.12a	26.3 ± 0.75a
		Pupal wt, mg	—	76.2 ± 4.57a	83.4 ± 6.32a	84.6 ± 4.56a	74.9 ± 4.96a	77.8 ± 5.13a	82.1 ± 2.37a
Perillaldehyde	199	% pupation	35.0 ± 5.00b	75.0 ± 5.00a	85.0 ± 15.0a	90.0 ± 5.77a	85.0 ± 9.57a	80.0 ± 8.16a	90.0 ± 10.0a
		% emergence	30.0 ± 5.77c	70.0 ± 5.77b	75.0 ± 12.6b	70.0 ± 5.77b	75.0 ± 9.51b	65.0 ± 5.00b	85.0 ± 5.00a
		Larval period, d	24.7 ± 0.83a	27.7 ± 1.12a	25.8 ± 0.91a	27.7 ± 1.75a	25.4 ± 0.79a	23.4 ± 0.75a	26.3 ± 0.75a
		Pupal wt, mg	94.0 ± 6.58a	81.7 ± 6.17b	84.1 ± 5.73b	86.9 ± 4.62b	79.8 ± 4.54b	74.6 ± 4.30b	82.1 ± 2.37b
Perillyl alcohol	179	% pupation	—	75.0 ± 9.57b	95.0 ± 5.00a	85.0 ± 5.00a	90.0 ± 5.77a	90.0 ± 5.77a	90.0 ± 10.0a
		% emergence	—	70.0 ± 12.9ab	75.0 ± 9.51ab	65.0 ± 12.6b	70.0 ± 10.0b	85.0 ± 9.51a	85.0 ± 5.00a
		Larval period, d	—	27.8 ± 1.45a	26.4 ± 1.40a	26.8 ± 1.30a	25.2 ± 1.08a	23.8 ± 0.98a	26.3 ± 0.75a
		Pupal wt, mg	—	88.3 ± 4.75a	83.2 ± 4.89a	77.0 ± 5.46a	78.6 ± 3.02a	91.2 ± 4.55a	82.1 ± 2.37a
α -Pinene	199	% pupation	60.0 ± 14.1b	90.0 ± 10.0a	90.0 ± 5.77a	85.0 ± 15.0a	65.0 ± 17.1b	75.0 ± 5.00ab	90.0 ± 10.0a
		% emergence	40.0 ± 11.5c	65.0 ± 5.00b	70.0 ± 10.0b	70.0 ± 19.1b	40.0 ± 18.3c	60.0 ± 8.16b	85.0 ± 5.00a
		Larval period, d	26.3 ± 1.10a	24.2 ± 0.78a	25.7 ± 0.75a	24.1 ± 0.74a	23.5 ± 0.77a	23.9 ± 0.87a	26.3 ± 0.75a
		Pupal wt, mg	81.3 ± 7.22a	73.2 ± 4.20ab	83.6 ± 2.95a	75.5 ± 5.02ab	61.1 ± 6.49b	73.5 ± 6.61ab	82.1 ± 2.37a
Pulegone	199	% pupation	0 ± 0b	0 ± 0b	80.0 ± 14.1a	85.0 ± 5.00a	95.0 ± 5.00a	85.0 ± 5.00a	90.0 ± 10.0a
		% emergence	0 ± 0b	0 ± 0b	65.0 ± 17.1a	70.0 ± 12.9a	80.0 ± 20.0a	75.0 ± 9.51a	85.0 ± 5.00a
		Larval period, d	—	—	25.9 ± 1.15a	22.1 ± 0.52a	23.4 ± 1.03a	21.8 ± 0.49a	26.3 ± 0.75a
		Pupal wt, mg	—	—	91.1 ± 5.45a	84.2 ± 4.44b	82.5 ± 4.14b	79.2 ± 4.20b	82.1 ± 2.37b
α -Terpineol	199	% pupation	35.0 ± 5.00c	65.0 ± 17.1b	95.0 ± 5.00a	80.0 ± 8.16a	80.0 ± 8.16a	85.0 ± 9.57a	90.0 ± 10.0a
		% emergence	5.00 ± 5.00b	65.0 ± 17.1a	75.0 ± 9.57a	70.0 ± 10.0a	65.0 ± 9.57a	75.0 ± 5.00a	85.0 ± 5.00a
		Larval period, d	31.3 ± 1.67a	24.0 ± 0.65b	24.3 ± 0.81b	24.3 ± 1.08b	24.8 ± 1.28b	23.2 ± 0.82b	26.3 ± 0.75b
		Pupal wt, mg	66.7 ± 6.35b	93.3 ± 4.56a	81.7 ± 4.82a	86.3 ± 3.60a	91.1 ± 4.70a	82.4 ± 3.81a	82.1 ± 2.37a

Table 4. Continued

Compound	<i>n</i> ^a	Development	Concentrations ^b						Control (blank)
			20 mg	2 mg	0.2 mg	0.02 mg	2 µg	0.2 µg	
Thujone	199	% pupation	0 ± 0c	73.3 ± 13.3b	100 ± 0a	80.0 ± 20.2b	100 ± 0a	100 ± 0a	90.0 ± 10.0a
		% emergence	0 ± 0c	66.7 ± 17.9b	86.7 ± 6.93a	66.7 ± 17.9b	93.3 ± 6.93a	80.0 ± 11.5a	85.0 ± 5.00a
		Larval period, d	—	25.4 ± 0.81a	24.6 ± 0.95a	25.3 ± 1.06a	24.8 ± 1.14a	23.4 ± 0.93a	26.3 ± 0.75a
Thymol	199	Pupal wt, mg	—	76.3 ± 4.25a	75.4 ± 5.02a	67.8 ± 6.81ab	76.0 ± 3.87a	56.2 ± 4.06b	82.1 ± 2.37a
		% pupation	0 ± 0c	50.0 ± 5.77b	85.0 ± 9.57a	85.0 ± 9.57a	75.0 ± 9.57ab	90.0 ± 5.77a	90.0 ± 10.0a
		% emergence	0 ± 0c	45.0 ± 5.00b	65.0 ± 17.1b	50.0 ± 5.77b	60.0 ± 14.1b	90.0 ± 5.77a	85.0 ± 5.00a
MTEE-25 ^c	179	Larval period, d	—	27.0 ± 2.14a	29.2 ± 1.52a	28.8 ± 1.74a	28.5 ± 1.57a	24.8 ± 1.13a	26.3 ± 0.75a
		Pupal wt, mg	—	85.2 ± 4.43a	88.4 ± 4.41a	84.9 ± 6.21a	79.2 ± 3.40a	87.2 ± 3.21a	82.1 ± 2.37a
		% pupation	—	0 ± 0c	35.0 ± 9.57b	55.0 ± 20.6b	55.0 ± 22.2b	60.0 ± 8.16b	90.0 ± 10.0a
MTEE-35	179	% emergence	—	0 ± 0c	20.0 ± 8.16b	25.0 ± 18.9b	25.0 ± 15.0b	25.0 ± 15.0b	85.0 ± 5.00a
		Larval period, d	—	—	28.6 ± 1.83a	29.9 ± 1.72a	27.9 ± 1.45a	24.2 ± 1.02a	26.3 ± 0.75a
		Pupal wt, mg	—	—	64.3 ± 6.88b	60.6 ± 5.06b	71.4 ± 5.25b	54.7 ± 5.60b	82.1 ± 2.37a
MTEE-76	179	% pupation	—	70.0 ± 10.0b	75.0 ± 9.57b	85.0 ± 9.57a	75.0 ± 5.00b	85.0 ± 5.00a	90.0 ± 10.0a
		% emergence	—	55.0 ± 20.6b	55.0 ± 17.1b	55.0 ± 22.2b	65.0 ± 17.1b	80.0 ± 8.16a	85.0 ± 5.00a
		Larval period, d	—	27.7 ± 1.48a	29.0 ± 1.11a	27.6 ± 1.27a	24.1 ± 0.83a	25.5 ± 1.08a	26.3 ± 0.75a
MTEE-90	199	Pupal wt, mg	—	83.7 ± 5.67a	83.3 ± 5.11a	78.0 ± 3.13a	82.6 ± 3.87a	85.7 ± 4.66a	82.1 ± 2.37a
		% pupation	—	15.0 ± 5.00c	35.0 ± 5.00c	70.0 ± 5.77b	90.0 ± 5.77a	85.0 ± 9.57a	90.0 ± 10.0a
		% emergence	—	15.0 ± 5.00b	30.0 ± 5.77b	60.0 ± 14.1a	75.0 ± 12.6a	65.0 ± 12.6a	85.0 ± 5.00a
MTEE-99	179	Larval period, d	—	32.3 ± 2.18a	26.9 ± 2.20b	24.9 ± 0.84b	25.9 ± 1.07b	25.4 ± 1.15b	26.3 ± 0.75b
		Pupal wt, mg	—	76.7 ± 2.89b	89.7 ± 11.1a	83.8 ± 3.53a	82.5 ± 3.49a	75.9 ± 3.84b	82.1 ± 2.37a
		% pupation	0 ± 0d	70.0 ± 10.0bc	75.0 ± 5.00b	80.0 ± 8.16b	75.0 ± 9.57b	65.0 ± 23.6c	90.0 ± 10.0a
MTEE-P	199	% emergence	0 ± 0c	60.0 ± 8.16b	65.0 ± 9.57b	85.0 ± 5.00a	65.0 ± 5.00b	55.0 ± 5.00b	85.0 ± 5.00a
		Larval period, d	—	28.4 ± 1.57a	23.8 ± 0.99a	26.3 ± 1.13a	24.7 ± 1.32a	26.2 ± 1.39a	26.3 ± 0.75a
		Pupal wt, mg	—	79.4 ± 4.76a	86.0 ± 5.03a	89.6 ± 4.08a	86.3 ± 5.86a	74.9 ± 5.82a	82.1 ± 2.37a
Pyrethrins ^d	199	% pupation	—	55.0 ± 9.57b	50.0 ± 12.9b	25.0 ± 5.00b	45.0 ± 17.1b	30.0 ± 12.9b	90.0 ± 10.0a
		% emergence	—	40.0 ± 8.16b	45.0 ± 15.0b	25.0 ± 5.00b	35.0 ± 12.6b	20.0 ± 8.16b	85.0 ± 5.00a
		Larval period, d	—	26.0 ± 1.66a	26.2 ± 1.26a	24.8 ± 0.73a	25.9 ± 1.17a	25.9 ± 1.53a	26.3 ± 0.75a
Rotenone	199	Pupal wt, mg	—	78.4 ± 6.09b	79.5 ± 4.81b	91.0 ± 0.01a	77.7 ± 6.53b	78.7 ± 0.01b	82.1 ± 2.37b
		% pupation	0 ± 0d	0 ± 0d	20.0 ± 8.16c	70.0 ± 5.77b	70.0 ± 5.77b	95.0 ± 5.00a	90.0 ± 10.0a
		% emergence	0 ± 0d	0 ± 0d	20.0 ± 8.16c	60.0 ± 8.16b	50.0 ± 5.77b	80.0 ± 8.16a	85.0 ± 5.00a
Pyrethrins ^d	199	Larval period, d	—	—	25.3 ± 0.75a	27.1 ± 1.26a	26.1 ± 1.39a	24.3 ± 0.85a	26.3 ± 0.75a
		Pupal wt, mg	—	—	82.8 ± 7.55a	87.4 ± 3.18a	78.4 ± 5.64a	82.9 ± 3.95a	82.1 ± 2.37a
		% pupation	0 ± 0c	0 ± 0c	0 ± 0c	0 ± 0c	0 ± 0c	60.0 ± 8.16b	90.0 ± 10.0a
Rotenone	199	% emergence	0 ± 0c	0 ± 0c	0 ± 0c	0 ± 0c	0 ± 0c	30.0 ± 10.0b	85.0 ± 5.00a
		Larval period, d	—	—	—	—	—	29.3 ± 2.32a	26.3 ± 0.75a
		Pupal wt, mg	—	—	—	—	—	64.9 ± 6.00b	82.1 ± 2.37a
Pyrethrins ^d	199	% pupation	0 ± 0c	0 ± 0c	0 ± 0c	20.0 ± 8.16b	70.0 ± 12.9a	85.0 ± 9.57a	90.0 ± 10.0a
		% emergence	0 ± 0c	0 ± 0c	0 ± 0c	15.0 ± 9.57b	40.0 ± 14.1b	70.0 ± 12.9a	85.0 ± 5.00a
		Larval period, d	—	—	—	33.8 ± 2.36a	29.4 ± 1.39a	26.1 ± 1.32a	26.3 ± 0.75a
Pyrethrins ^d	199	Pupal wt, mg	—	—	—	76.5 ± 6.05a	71.1 ± 5.40ab	66.5 ± 5.48b	82.1 ± 2.37a

All values are means ± standard error of mean (SEM) of replicates. Means within a row followed by the same letter are not significantly different (LSD, *P* = 0.05; SAS Institute 1991).

^a Number of individual *O. nubilalis* larvae tested.

^b Concentrations are expressed in mg or µg of compounds on the diet in cups.

^c MTEE—Monoterpenoid derivatives (see Fig. 2).

^d Concentration adjusted for 20% (AI).

rimentially at dietary concentrations as low as 10 ppm, whereas neonates were unaffected at 1,000 ppm. The older larvae may have had difficulty adapting to the *l*-menthol-treated diet, as reflected in survival and length of the larvae development period. In both developmental toxicity tests, beginning with either 1st or 2nd instars, rotenone was more effective than the monoterpenoids in increasing mortality or the length of the larval and pupal development periods or decreasing the percentage pupation and emergence.

During these experiments, some larvae on the diet treated with monoterpenoids crawled on the lids or walls of the cup, avoiding contact with the diet, and no feeding was evident. Death often occurred for those larvae before molting to the next instar. This indicated that some monoterpenoid compounds may have antifeedant or repellent effects; consequently, some larvae might be dead by starvation as well as by contact toxicity. Further study will be required to determine the major reasons for larval mortality in comparison

Table 5. Effects of selected monoterpenoids incorporated into the diet on development of neonate *O. nubilalis*, beginning with neonate 1st instars

Compound	n ^a	Development	Concentration, ppm ^b					Control 1 (blank)	Control 2 (acetone)
			1,000	100	10	1	0.1	0.01 ppm	
l-Menthol	240	% pupation	90.0 ± 6.83a	90.0 ± 4.47a	93.3 ± 4.22a	93.3 ± 6.15a	96.7 ± 3.39a	96.7 ± 3.39a	90.0 ± 4.47a
		% emergence	53.3 ± 6.67a	53.3 ± 6.67a	63.3 ± 12.0a	63.3 ± 6.15a	60.0 ± 7.31a	66.7 ± 9.89a	56.7 ± 9.85a
		Larval period, d	30.0 ± 0.47a	29.4 ± 0.25a	29.4 ± 0.45a	29.5 ± 0.43a	29.5 ± 0.32a	28.5 ± 0.51a	27.6 ± 0.30a
		Pupal period, d	12.0 ± 0.68a	10.1 ± 0.30a	11.2 ± 0.48a	11.3 ± 0.28a	10.7 ± 0.29a	10.6 ± 0.27a	10.4 ± 0.23a
		Pupal wt, mg	76 ± 3.8a	76 ± 3.1a	77 ± 3.9a	76 ± 3.6a	76 ± 4.3a	82 ± 3.2a	70 ± 4.3a
Pulegone	240	% pupation	0 ± 0d	56.7 ± 9.55c	73.3 ± 8.44bc	86.7 ± 6.67ab	86.7 ± 4.22ab	93.3 ± 6.83a	90.0 ± 4.47ab
		% emergence	0 ± 0c	33.3 ± 8.44b	46.7 ± 8.44ab	70.0 ± 11.3a	66.7 ± 8.44a	56.7 ± 8.03ab	56.7 ± 9.85ab
		Larval period, d	—	29.9 ± 0.44a	29.8 ± 0.99a	29.2 ± 0.53a	29.2 ± 0.38a	29.8 ± 0.55a	27.6 ± 0.30a
		Pupal period, d	—	11.5 ± 1.46a	11.3 ± 0.65a	10.3 ± 0.27a	11.1 ± 0.33a	10.8 ± 0.47a	10.4 ± 0.23a
		Pupal wt, mg	—	63 ± 3.2b	74 ± 4.7a	76 ± 3.7a	74 ± 4.2a	69 ± 3.6a	70 ± 4.3a
Rotenone	240	% pupation	0 ± 0c	0 ± 0c	0 ± 0c	26.7 ± 6.67b	90.0 ± 4.47a	96.7 ± 3.39a	90.0 ± 4.47a
		% emergence	0 ± 0c	0 ± 0c	0 ± 0c	6.70 ± 4.22c	40.7 ± 11.2b	73.3 ± 8.44a	56.7 ± 9.85ab
		Larval period, d	—	—	—	44.0 ± 1.70a	32.4 ± 0.57b	29.4 ± 0.35b	27.6 ± 0.30b
		Pupal period, d	—	—	—	14.0 ± 0a	12.2 ± 0.80a	10.5 ± 0.25b	10.4 ± 0.23b
		Pupal wt, mg	—	—	—	59 ± 6.4b	75 ± 4.2a	69 ± 3.0a	70 ± 4.3a

All values are means ± standard error of mean (SEM) of 6 replicates. Means within a row followed by the same letter are not significantly different (LSD, P = 0.05; SAS Institute 1991).

^a Number of individual *O. nubilalis* larvae tested.

^b Concentrations are expressed as milligrams of compounds per gram of diet.

with the correlation of antifeedant activity or repellency. Although these larvicidal toxicity data suggest that some monoterpenoids had some degree of insecticidal activity and some of the synthetic derivatives showed enhanced activity, these monoterpenoids are less effective than pyrethrins and rotenone, the 2 natural insecticides used as standards.

The chronic evaluation study investigated the insect growth inhibitory effects of monoterpenoid compounds. Some monoterpenoid compounds adversely influenced growth and development of the European corn borer. Pupation rates were affected by *l*-menthol (20–30%) at all concentrations tested, and some monoterpenoid derivatives (MTEE-25, -99, -P) showed significantly reduced pupation rates (20–70% compared with controls). Pulegone also reduced pupation rate. Emergence rates were relatively lower than the untreated diet. MTEE-25 significantly reduced emergence rates (25–30%) compared with the untreated control. However, none of the treatments significantly affected durations of the pupal period, although geraniol, *l*-menthol, and pulegone delayed larval periods of 2nd-instar European corn borers in the in-diet test. Furthermore, in most monoterpenoid compound treatments, pupal weight was reduced at 1 or more concentrations.

Harwood et al. (1990) reported that menthol, menthone, and pulegone deleteriously affected in growth, feeding, and pupation of *Peridroma saucia* (Hübner) larvae. Pulegone is an effective defensive monoterpenoid because of its antifeedant activity that also affects insect development and reproduction (Gunderson et al. 1985). *d*-Limonene, linalool, β -myrcene, and α -terpineol significantly reduced the days required for development through the nymphal stages in the German cockroach, *Blattella germanica* (L.) fed treated diet, and especially *d*-limonene significantly shortened the time required to reach the adult (Karr and Coats 1992). Two monoterpenoids also have been shown to reduce the development time for *Aedes aegypti* mosquito larvae (Tsao et al. 1995). Binder et al. (1995) reported that structural modification at a single terminal functional group in the plant sesquiterpenoids farnesene, nerolidol, and farnesol affected the ovipositional behavior of European corn borer females. Beninger et al. (1993) reported in an artificial diet study that diterpenes, from another terpenoid group, also showed the potential to protect crops from the European corn borer; as an example, 3-epicaryoptin increased mortality and reduced growth of the European corn borer.

In summary, larvicidal and insect growth regulation activity could be beneficial as possible control agents for the European corn borer. Some natural monoterpenoids showed larvicidal activity and insect growth regulation effects in the current study, and derivatization created some compounds (2-fluoroethyl thymyl ether and propargyl citronellate) with enhanced biological activity against the European corn borer; however, they are not as potent as many commercial insecticides. Further research is needed to synthesize more enhanced derivatives and to elucidate the mech-

Table 6. Effects of selected monoterpenoids incorporated into the diet on development of *O. nubilalis*, beginning with late 2nd instars

Compound	n ^a	Development	Concentration, ppm ^b					Control 1 (blank)	Control 2 (acetone)
			10,000	1,000	100	10	1 ppm		
Geraniol	160	% pupation	50.0 ± 12.9b	75.0 ± 12.6ab	95.0 ± 5.00a	90.0 ± 5.8a	60.0 ± 8.2b	93.3 ± 5.17a	93.3 ± 5.17a
		% emergence	10.0 ± 5.78b	40.0 ± 8.17ab	45.0 ± 5.00ab	50.0 ± 23.8a	35.0 ± 9.58ab	53.3 ± 10.33a	53.3 ± 10.33a
		Larval period, d	26.9 ± 1.40a	16.1 ± 0.50bc	18.1 ± 0.46b	16.7 ± 0.48bc	19.1 ± 0.66b	14.9 ± 0.35c	15.2 ± 0.48c
		Pupal period, d	9.50 ± 5.50a	11.6 ± 0.33a	12.6 ± 0.50a	11.3 ± 0.55a	11.3 ± 0.51a	11.3 ± 0.31a	11.3 ± 0.49a
		Pupal wt, mg	73 ± 4.5c	85 ± 4.9b	93 ± 4.6a	83 ± 3.3b	96 ± 4.0a	87 ± 2.3b	82 ± 4.2b
l-Menthhol	160	% pupation	0 ± 0c	30.0 ± 5.78b	30.0 ± 10.0b	40.0 ± 8.17b	70.0 ± 17.3a	93.3 ± 5.17a	93.3 ± 5.17a
		% emergence	0 ± 0c	20.0 ± 11.6bc	20.0 ± 8.17bc	15.0 ± 9.58bc	45.0 ± 15.0ab	53.3 ± 10.33a	53.3 ± 10.33a
		Larval period, d	—	22.3 ± 3.02b	24.2 ± 2.75b	29.1 ± 2.55a	19.43 ± 1.00b	14.9 ± 0.35c	15.2 ± 0.48c
		Pupal period, d	—	11.0 ± 1.08a	11.5 ± 1.66a	13.0 ± 1.00a	11.1 ± 1.10a	11.3 ± 0.31a	11.3 ± 0.49a
		Pupal wt, mg	—	93 ± 5.8a	76 ± 4.0b	69 ± 6.5b	86 ± 3.7a	87 ± 2.3a	82 ± 4.2a
Pulegone	160	% pupation	0 ± 0c	0 ± 0c	65.0 ± 5.00b	65.0 ± 9.58b	55.0 ± 15.0b	93.3 ± 5.17a	93.3 ± 5.17a
		% emergence	0 ± 0c	0 ± 0c	35.0 ± 5.00b	25.0 ± 9.58b	30.0 ± 5.78b	53.3 ± 10.33a	53.3 ± 10.33a
		Larval period, d	—	—	21.9 ± 1.49b	19.8 ± 0.82b	28.3 ± 1.43a	14.9 ± 0.35c	15.2 ± 0.48c
		Pupal period, d	—	—	12.3 ± 1.02a	12.6 ± 0.40a	11.7 ± 0.49a	11.3 ± 0.31a	11.3 ± 0.49a
		Pupal wt, mg	—	—	81 ± 4.6a	85 ± 5.3a	58 ± 3.5b	87 ± 2.3a	82 ± 4.2a
Rotenone	160	% pupation	0 ± 0c	0 ± 0c	0 ± 0c	65.0 ± 9.58b	70.0 ± 12.9b	93.3 ± 5.17a	93.3 ± 5.17a
		% emergence	0 ± 0c	0 ± 0c	0 ± 0c	30.0 ± 5.78b	55.0 ± 15.0a	53.3 ± 10.33a	53.3 ± 10.33a
		Larval period, d	—	—	—	28.4 ± 1.72a	18.6 ± 0.75b	14.9 ± 0.35c	15.2 ± 0.48c
		Pupal period, d	—	—	—	11.0 ± 0.95a	12.2 ± 0.42a	11.3 ± 0.31a	11.3 ± 0.49a
		Pupal wt, mg	—	—	—	67 ± 4.1b	92 ± 5.9a	87 ± 2.3a	82 ± 4.2a

All values are means ± standard error of mean (SEM) of 6 replicates. Means within a row followed by the same letter are not significantly different (LSD, P = 0.05; SAS Institute 1991).
^a Number of individual *O. nubilalis* larvae tested.
^b Concentrations are expressed as milligrams of compounds per gram of diet.

anisms of regulation on insect growth and development induced by monoterpenoids. We hope this study will contribute toward the consideration of monoterpenoids for natural means of insect control and toward understanding the modes of biological action of the monoterpenoids.

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